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Artificial diet development [for abalone]

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Artificial diet development

[The following is based on a review paper entitled *The development of artificial diets for abalone* by AE Fleming, RJ van Barneveld and PW Hone that was published in **Aquaculture** 140 (1996): 5-53. Our writer **Eva Aldon** has simplified the presentation and deleted the citations. Full references can be found in the original paper. - *Ed.*]

The search for artificial diet for abalone aquaculture started some 30 years ago, and has intensified in recent years. To date, 28 research groups from around the world are trying to develop artificial diets. Japan has six such groups, including Nihon Nosan Kogyo KK which is probably the world's leading feed company. [The company sold 300 tons of abalone artificial feeds in 1993.] Australia and China have four research groups each; the United States, three; New Zealand, two; South Africa, three; Canada, Mexico, France, Korea, Thailand, Ireland, one each. The Philippines and Taiwan -- both with commercial fishery and taking fledgling steps towards establishing abalone aquaculture - do not have any yet. The studies at AQD, for instance, are still focused on induced spawning and larval rearing although cage farming studies have been initiated. A lone study on artificial feeds and feeding is being conducted by AQD scientists Myrna Teruel and Oseni Millamena. Abalone is just one of the 20 or so commodities prioritized for research at AQD from 1998 to year 2000.

Existing artificial diets

The artificial abalone diets in the market are similar in their proximate composition, as can be seen below:

	Range	*Average*
Protein	20-50%	30%
Carbohydrate	30-60%	47%
Lipid	1.5-5.3%	4%
Crude fiber	0-3%	
Moisture	-	12%

The capacity for abalone to digest fiber is limited. The growth of *Haliotis discus hannai* decreased as cellulose content of the diet was increased from 0 to 20%. This suggests that abalone have a poor ability to digest cellulose, despite the presence of cellulases in the gut. Some artificial diets contain fiber for binding purposes, hence the level of fiber can be as much as 6% of dry weight.

Protein sources and optimal inclusion levels

To meet the protein needs of abalone is to establish specific amino acid requirements, the most appropriate balance of dietary amino acids, and the availability of amino acids from a range of protein sources.

It was reported that incorporating fishmeal to formulate diets containing 27 and 32% crude protein produced a similar monthly growth rate of 32% of body weight. A variety of protein sources (with 30% protein content) was tested for 3-5 g abalone at 20°C.

The most commonly used protein sources in abalone diets are fishmeal, defatted soybean meal and casein. Fishmeal, however, is used extensively in aquaculture feeds, hence the high demand worldwide. But there's been a worldwide decline of fishmeal production which may increase cost. It should be noted that diets containing high levels of fishmeal are detrimental to the environment because they contain excessive amounts of phosphorus. Soybean is a potential replacement to fishmeal because its amino acid profile is close to that of fish, and its protein is highly digestible.

Mackerel silage and abalone viscera silage are cheap protein sources and reportedly gave good growth rates with a diet consisting of 20% silage, 16.8% fishmeal and 10% soybean meal. Abalone viscera silage is also a potential to enhance digestion.

All essential amino acids are available in a synthetic form and are important for supplementation in the diet. Protein quality may be improved by matching the amounts of amino acids in the diet with those in the body. The diet with DL-methionine was formulated

using the amino acid ratios in the body rather the absolute values.

Energy / carbohydrate sources

Abalone consume a natural diet consisting of 40-50% carbohydrate and have various enzymes capable of hydrolysing complex carbohydrates. Cheap cereal sources, such as wheat and corn flour, soybean meal, maize or rice starch are frequently used as an energy source. Starches can be both an energy source and a binder in many commercial abalone feeds. The metabolic rate of abalone is low, hence, energy is also low. The energy content of feeds exceeding the requirement for tissue synthesis may be converted into glycogen. Excessive energy in the diet may lead to poor utilization of the protein which reduces feeding thus energy is wasted.

Vitamins and minerals

The requirement by abalone for vitamins and minerals were based on the requirements for carp and rainbow trout. The optimum level of the mineral mix in the diet was determined by adding graded levels between 0 and 16% into a test diet and gave a maximum growth at 8% and later 4% to improve solubility of the pellets. The effect of adding Ca and P to artificial diets was also investigated. Vitamin C is an important component of the diet.

Feed stimulants and attractants

Feed stimulants such as algae and seaweeds are added to the diet to enhance food intake and growth rate but reportedly has no effect in consumption. Abalone consumed more when supplemented with Taremel A40 while using dried kelp in diets was found unnecessary. Fishmeal is found to be more palatable than a casein diet. The leaching of soluble proteins and free amino acids from abalone viscera and fish silage attracts abalone to the feed but this was not an effective stimulant like soybean, fishmeal and kelp meals.

Palatable dietary ingredients added to the diet to enhance intake is a practical and economically viable option. Fishmeal, added as a protein source, was found to enhance intake more than a casein diet. A combination of ingredients with attraction and palatibility will improve the acceptability of the feed.

There are extensive studies on the feeding attractants of *Haliotis discus:* algae, chemical fractions extracted from algae, whole proteins, nucleic acid-related compounds, amino acids and peptides and combinations of amino acids. Combinations of some non-volatile nitrogenous bases, amino acids (as peptides) and proteins are generally more effective and attractive to abalone than when present individually. The acrid spices are potential for artificial diets as they are may be an attractant and stimulant in addition to having antiseptic properties.

Practical feed stimulants or attractants for artificial feeds may be readily identified by using feed ingredients containing high concentrations of the amino acid and fatty acid group of compounds.

Binders

Aquatic animal feeds require a binder to keep the feed intact for at least 2 days in water. Being crucial to the development of a successful feed for abalone, binders in artificial feeds are the most guarded of information and are often patented.

The majority of commercial companies use starches and gluten for binders. Although gels are frequently used to bind experimental diets, they are not considered economically viable for commercial feeds. Water solubility of a mixture of alginates and flours (rice starch, sodium alginate and gelatin) as binders were compared. A binding technique using starches and alginates was developed in Australia.

Feed stability of existing artificial diets

Pellet stability refers to the stability of the binder when immersed in seawater and is dependent on the binding ingredients, particle size and manufacturing process. The average stability of abalone feeds is about 2-3 days. A combination of agar and gelatin (1:3) forming 20% of the diet improved the stability of the feed so that only 20% was lost after 6 hours and 30% after 24 h. Further manipulations of these ratios and concentrations in the diet may improve water stability while lowering cost.

Feed stability should be tested in conditions that simulate commercial temperature, aeration and flow regimes.

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Leaching of nutrients and microencapsulation

Leaching is the loss of nutrients from the feed against the loss or breaking up of the binders which may occur without a change in feed weight. Leaching, recognized as a problem by feed formulators, can be controlled by microencapsulation, the binding technique, and heat treatment during manufacture process. Microencapsulation is a technique of enclosing dietary nutrients within a digestible capsule wall to reduce leaching and bacterial degradation. The capacity of abalone to digest the cellulose coating depends on the species.

Feed decomposition

The use of artificial feeds in culture systems causes feed decomposition and the subsequent deterioration of the water quality. Decomposition may be due to the susceptibility of the meal protein to bacterial attack or oxidation of the lipids in the meal. Inclusion of natural antiseptic substances may slow the rate of decomposition.

Feed availability in the culture system

The farmers are more concerned with accessibility of feed to animals in the tanks than nutritive quality and recognized that tank design is important to achieve feed availability and water quality. Ideally a feed should meet the following requirements:

- that the amount of uneaten feed is minimized
- that uneaten feed does not pollute the tank
- that animals are feeding to satiation
- that animals do not expend significant amounts of energy searching for feed
- that feed and faeces do not occur in the same place in the tank

Artificial feeds are buoyant to simulate the algae in the culture tank. Not all abalone will readily search for food each night hence heavier material is used during the day and lighter material at night.

Abalone performance when fed artificial diets

Intake is the product of feeding rate and duration of feeding and is different from the rate of feeding. Temperature and the light regime show a relationship between intake and the duration of darkness. The required amount of feed added to the tank to maximize growth rate is approximately twice the daily intake.

The quantity of food the abalone consumes each day varies depending on their metabolic liveweight. The amount of energy in artificial feeds is extremely high compared with the daily energy requirements of abalone, thus, abalone may cease feeding before they have consumed adequate quantities of nutrients.

Growth rate

It is difficult to compare the growth performance of abalone on various diets as abalone species differ in their capacity for growth. If a diet is deficient in a nutrient, the growth rate may decrease or cease after the abalone have exhausted its own stores.

Ideally, the temperature should be kept constant during trials or temperature data should be reported with the growth rate to determine if temperature is a factor.

Nutrient requirements of abalone for use in the formulation of artificial diets

The artificial diet of abalone should match its diet requirement for maximum efficiency and profit and minimum ecological impact. The artificial diets in the market can still be improved in terms of formulation and efficiency.

The difficulties in determining the nutrient requirements of abalone include:

- maintaining an experimental diet underwater with minimal nutrient loss
- difficulty associated with measuring slow growth rates
- the long duration of experiments
- absorption of nutrients by abalone from the aquatic environment
- maintenance of a constant experimental environment

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The first consideration when formulating feeds is to ensure that the essential amino acids and energy are supplied in sufficient quantities and proportions to meet the requirements of the animal.

Response of abalone to protein and energy intake

The first consideration when formulating feeds is to ensure that the essential amino acids and energy are supplied in sufficient quantities. Both amino acids and energy are closely interlinked in growth and development. If there is insufficient energy, the surplus amino acids (they cannot be stored) are wasted. If there is insufficient amino acid relative to energy, then the development of lean tissue is restricted. No work has yet been done to determine the optimum ratio of amino acids to energy in diets for abalone. The most valuable information in relation to the protein and energy requirements of abalone could be met by investigating the interactive effects of protein and energy intakes on protein deposition and the relationship between energy intake and protein deposition to establish a protein:energy ratio for use in diet formulations.

There are three areas that affect protein utilization:

- the quantity or proportion of dietary protein
- the quality or amino acid make-up of the dietary protein
- the ability of the abalone to utilize non-protein dietary components as energy sources

Utilizing non-protein sources is being studied in terms of absorption, storage and expenditures of energy in abalone diets with different ratios of protein, lipid and carbohydrates.

Amino acid requirements

Proteins are complex compounds of mixtures of amino acids forming various proteins in the muscles, organs, and secretions such as enzymes and hormones.

Further research is needed to establish specific amino acid requirements, the most appropriate balance of dietary amino acids, and the availability of amino acids from a range of protein sources. Most protein contain 19 or more amino acids, of which nine are essential and must be supplied in the diet.

Ideal protein ratio

A knowledge of amino acid requirements could improve the efficiency of protein use in artificial abalone diets. The concentration of amino acids in the soft tissue of abalone could indicate the balance of amino acids required in the diet.

Lysine may be the major limiting amino acid in abalone when fed a cereal-based diet.

Energy requirements

Meeting the energy requirements of abalone includes these considerations: (1) the system should be precise and simple to apply; (2) the values should be additive; and (3) the values of the feed can be easily estimated. Energy requirement is dependent on temperature and body weight.

Lipid requirements

The lipids are heterogenous group of compounds important to the diet because of their high energy value and because they are sources of essential fatty acids and fat-soluble vitamins. Fatty acids of abalone are significantly different from other animals due to differences in diet composition. The abalone's lipid requirement is very low although the abalone is highly efficient in utilizing lipid.

Vitamin and mineral requirements

There are a few studies on vitamin requirements of abalone. Digestive bacteria may significantly contribute to the vitamin nutrition of abalone implying oversupply of these nutrients.

The requirements of the juveniles

The capacity of juvenile abalone to digest various nutrients may vary according to age and size. There may be nutritional differences between juvenile and adult abalone although no comparative investigations have been done. Juveniles generally require more pro-

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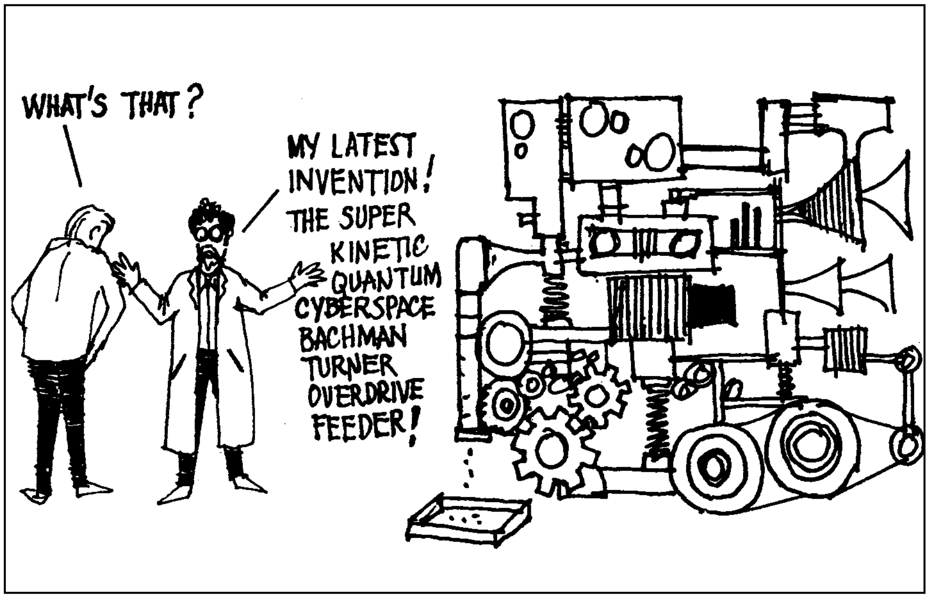

since little was understood of the culture practices. It is only now that institutional effort is being made to save the shrimp industry but it is already quite late. High density milkfish culture is the new craze and this can very well follow the footsteps of the shrimp industry. I think by being abreast with industry, scientists are in a better position to detect early on production problems before these happen. If problems do indeed happen, rehabilitation at least will be easier. Whether we like it or not, entrepreneurs will invest where there is money. They will try to generate technology on their own if this is not available and this is not a good idea.

What do you think is the future direction of our aquaculture industry? Does the shrimp industry have a chance of being rehabilitated?

I think more and more aquaculture activities is going to be directed to marine cage and pen farming where there is large area available for expansion. We see this now for milkfish culture in Pangasinan, Davao, Leyte, Cebu, Iloilo, and Negros. Cage and pen farming is much more productive than pond culture and there is no need to spend for pumping or aeration. It is very attractive to investors. As for the shrimp industry, if the disease problems are controlled, there are still many farmers in Negros and other parts of the country that would want to come back. I am not so optimistic however because the intensification of milkfish is going to lower water quality in the coming years.

What are your plans?

I am focusing attention on cage farming. I am now experimenting on culture methodologies and all-weather cage facilities. Of course this will include the use of automatic feeders. There is much R&D I'd like to do and intend to do. My approach is not to reinvent the wheel but to adopt existing technologies to suit local farming needs and conditions, much like what I did with the Kinetic Feeder.

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tein and energy per body weight than adults having higher growth rate. Also, small juveniles may require a different amino acid balance because of different growth requirement as viscera and physiological processes develop. Adults may require more lipid in the diet during gonad development. Bacterial growth which develop in the surface of the feed during prolonged storage may even improve the feed's nutritional quality. Viable bacteria in the gut of juveniles may also contribute significant quantities of nutrients to the diet and could perform metabolic activities in the gut that are highly significant to the abalone's development. Strains of these bacteria are capable of hydrolysing a variety of complex polysaccharides in algae.

Artificial diet enhances growth of hatchery-stage juveniles. It can improve survival and can aid in broodstock management. Artificial diets can be cost-effective since these improve productivity.

Nutritional value of ingredients

The best way to establish the nutritional value of ingredients for use in artificial diets for abalone is to determine the availability of the nutrients within the ingredients, i.e. the proportion of nutrients capable of being used by the abalone.

Digestive enzymes and the digestive capacity of abalone

The digestive enzymes of abalone can be purchased commercially. These are used extensively to lyse cell walls so that the protoplasts can be extracted for experimental use. Proteolytic activity of *Haliotis discus* was most active in an acidic environment. The carbohydrases have been investigated for abalone species. The abundant and com-

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plex composition of the polysaccharide hydrolases in these species reflects the widespread occurrence in the brown algae the abalone feed on. Cellulase is found in the gut of a number of abalone species although they differ in the levels of activity. Abalone could not easily digest cellulose but its enzymes could efficiently digest the cell walls of red and brown algae. Pre-fed algae were more efficiently digested.

Abalone can alter their enzyme composition to cope with changes in their diet. Juvenile *H. midae* fed an artificial diet had higher protease and lower amylase levels in their gut compared with abalone fed diatoms, suggesting a potential energy and protein source. The similar level of lipase between the dietary groups indicates that lipids do not play an important role in the energy metabolism of abalone.

Digestibility trials

The digestibility of a food is defined as the proportion which is absorbed by the abalone and not excreted in the feces. Feces collection is impossible because they tend to disintegrate in the water, making it difficult to determine the digestibility of specific nutrients. Temperature and the light regime should be controlled as these affect digestibility.

Digestibility of the ingredients

The general observation for *Haliotis rubra, H. rufescens* and *H. laevigata* was that these can digest lipids with high efficiency. Reports on digestibility of energy in a fishmeal-based diet was about 48% for *H. rubra* and 43% for *H. laevigata,* however, this is lower than the digestibility for algae. Digestibility investigations are still required to further improve current artificial diets.

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